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Intramolecular Activation Mechanism of the Dictyostelium LRRK2 Homolog Roco Protein GbpC

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Supplementary Information

Primer	Sequence (5'-3')	Restriction sites	Amino acid coverage
Fw1	GGGATCC AAAAAAAT <u>GT</u> TCACAAAAACAAAAACCAAT	<i>Bam</i> HI	1-524
Rv1	<u>GTTATCTAGATGGTAC</u> CTTCTCTTTGAGATAAGGTTGAG	<i>Kpn</i> I, <i>Xba</i> I	
Fw2	GGGTACCA AGTAGTTTCTTTACACTC	<i>Kpn</i> I	522-868
Rv2	GGATATCT ACCATTGAAATATCTG	<i>Eco</i> RV	
Fw3	GGGATCC AAAAAT <u>GGATAT</u> CCCAATACCAAGATTTAATTTAAG	<i>Bam</i> HI, <i>Eco</i> RV	867-1160
Rv3	<u>GTTATCTAGAACCTAGG</u> ATTTGACAATACTTGTAAG	<i>Avr</i> II, <i>Xba</i> I	
Fw4	GCCTAGGT CAAATTAAAAAGAAATTTGC	<i>Avr</i> II	1159-1411
Rv4	GTGGTACCG CATTTTACCCGCTGTATCTGAT	<i>Kpn</i> I	
Fw5	GAGATCT AAAAAT <u>GGCGGTACC</u> ACCACATTGGAGAAT	<i>Bgl</i> II, <i>Kpn</i> I	1408-1615
Rv5	<u>GTTATCTAGAGTCGCG</u> AGATTTAAAAATACTCTTGATAC	<i>Nru</i> I, <i>Xba</i> I	
Fw6	GAGATCT AAAAAT <u>GCTCGCG</u> ACTGGTATGAGAGATCCATT	<i>Bgl</i> II, <i>Nru</i> I	1613-2008
Rv6	<u>GTTATCTAGAACTCGAG</u> GTCAAACCACCACCATTCTC	<i>Xho</i> I, <i>Xba</i> I	
Fw7	GAGATCT AAAAAT <u>GACCTCGAG</u> TAATTTCTTTGGTAATGGTTC	<i>Bgl</i> II, <i>Xho</i> I	2005-2334
Rv7	<u>GTTATCTAGAGTCGAC</u> GTTTGAGTACGACCTAAATAAG	<i>Sal</i> I, <i>Xba</i> I	
Fw8	GAGATCT AAAAAT <u>GACGTCG</u> ACTTCACCATTGAATGAAGG	<i>Bgl</i> II, <i>Sal</i> I	2331-2631
Rv8A	GTCTAGATT AAGCATAAAGTTGTGATTCT	<i>Xba</i> I	
Rv8B	GTCTAGAA GCATAAAGTTGTGATTCTCT	<i>Xba</i> I	

SI Table I: Cloning of *gbpC* cDNA in *Dictyostelium* expression vectors.

gbpC cDNA was amplified with PCR from cDNA in eight parts of approximately equal size. Part 8 was amplified in two versions that differ in the presence of a Stop codon, to discriminate for expression of GbpC, and a GbpC-GFP fusion construct. Sequences in red represent restriction sites for direct cloning of separate parts in MB74-expression plasmids, sequences in bold represent restriction sites for fusion of parts, and Start/Stop codons are underlined.